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(54) Title: INHIBITION OF MICROBIAL GROWTH IN AQUEOUS STREAMS (57) Abstract The invention is a process for preventing microbial growth in aqueous streams by applying peracetic acid to the aqueous stream. Generally, the process of the invention is applicable to aqueous streams used in any number of applications such as the application of streams for the transport of fruits or vegetables into the processing environment and through the various steps of processing.		

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INHIBITION OF MICROBIAL GROWTH IN AQUEOUS STREAMS

Field of the Invention

5 The invention relates to the control of microbial growth in aqueous streams. More specifically, the invention relates to the control of microbial growth in aqueous streams used for transporting food products in processing environments such as fruit, vegetable and
10 food products, for example, mushrooms, poultry, tomatoes, and the like.

Background of the Invention

 The advent of food processing has long since
15 revolutionized both the availability of foods as well as the expectation of consumers for a large variety of high quality food products. Initially, food processing techniques included canning, and later, refrigeration, freezing, freeze drying as well as vacuum packaging.
20 The application of various constituent-based and process-based preservation systems have all lead to a wider availability of high quality food stuffs.

 In turn, food pricing and availability is generally subject to various constraints including environmental
25 hazards as well as natural weather cycles, selection and processing considerations, and overall economic and marketing constraints. Given the large volume of food selected and processed on an annual basis, as well as the relative uncontrollability of factors such as the
30 environment and the marketplace, producers strive to economize in the selection and processing of food stuffs. One means of processing a large volume of foods, such as, for example, fruits and vegetables, is after selection, to transport these various food stuffs
35 by an aqueous medium to deliver the food stuffs through various processing steps and environments.

 For example, in specific applications, fresh fruits and vegetables may be transported through water streams by food handling equipment used at the processing plant.

After picking, fruits and vegetables are introduced into a flume system wherein water acts as a transport medium and a cleaning medium. Water may be used to support and transport the fruits or vegetables from an unloading sight to a final storage or packing or processing location. During the transport, water can take a food item from an initial location through a series of somewhat separate stages to a final station where the produce is removed from the water and packed. The water within each stage may have a varying degree of organic load in the form of any number of sediments and soluble materials. This water is generally recycled.

Given the nature of the food transported as well as the presence of sediments and soluble materials, the water, flume, and other transport or processing equipment may be subject to the growth of unwanted microorganisms. These microorganisms are generally undesirable to the transported food, the water, the flume and may cause buildup on all water contact surfaces of slime or biofilm, which requires frequent cleaning to remove. Further, because the process water and equipment are in contact with food products, the control of unwanted microorganisms presents certain problems created by a food contact environment containing microorganisms.

Ideally, an antimicrobial agent or compound used in such a system will have several important properties in addition to its antimicrobial efficacy. The compound or agent should have no residual antimicrobial activity on the food. Residual activity implies the presence of a film of antimicrobial material which will continue to have antimicrobial effect which may require further rinsing of the food product. The antimicrobial agent preferably should also be odor free to prevent transfer of undesirable odors onto food stuffs. The antimicrobial agent should also be composed of direct food additive materials which will not affect food if

contamination occurs, nor effect humans should incidental ingestion result. In addition, the antimicrobial agent should preferably be composed of naturally occurring or innocuous ingredients, which are
5 chemically compatible with the environment and cause no concerns for toxic residues within the flume water.

The use of other antimicrobial agents in the control of microorganisms is well known for various applications. For example, Grosse Böwing et al U.S.
10 Patent Nos. 4,051,058 and 4,051,059 used peracetic acid as a food grade sanitizer in a variety of applications. Further, Greenspan et al, U.S. Patent No. 2,512,640 teach the use of a peracetic acid composition comprising
15 500 ppm or more of peracetic acid for the treatment of various fruit and vegetable compositions in a spray applicator. Greenspan et al, Food Technology, Vol. 5, No. 3, 1951, similarly discloses spray compositions which may be applied to fresh fruits and vegetables comprising peracetic acid. Langford, UK Patent
20 Application GB 2 187 958 A discloses the use of peracetic acid and propionic acid for the treatment of fungi in microbial plant pathogens on growing plants and especially edible crops.

In other publications, Baldry et al, "Disinfection
25 of Sewage Effluent with Peracetic Acid", Wat. Sci. Tech., Vol. 21, No. 3, pp. 203-206, 1989; and Poffé et al, "Disinfection of Effluents from Municipal Sewage Treatment Plants with Peroxy Acids", Zbl. Bakt. Hyg. I. Abt. Orig. B 167, 337-346, (1978) both disclose the use
30 of peroxy acids for the treatment of effluents streams and municipal sewage applications. Hutchings et al, "Comparative Evaluation of the Bactericidal Efficiency of Peracetic Acid, Quaternaries, and Chlorine-Containing Compounds", Society of American Bacteriologists,
35 Abstracts of Papers Presented at the 49th General Meeting, discloses the generally efficacy of peracetic acid compared to various other antimicrobial compounds.

Additionally, Branner-Jorgensen et al, U.S. Patent No. 4,591,565 discloses the reduction of the thermal stability of rennet through the use of aqueous-based aliphatic or inorganic peroxy acids. Block,
5 "Disinfection, Sterilization, and Preservation", Fourth Edition, Chapter 9, pages 167-181, discloses the various characteristics and attributes of peroxygen compounds. However, generally the art has taught against the use of percarboxylic acids in aqueous streams due to concerns
10 of compound stability in the presence of high concentrations of organic matter.

In the past, flume apparatus have generally been treated with sodium hypochlorite and chlorine dioxide. Generally, these materials are effective in preventing
15 the unwanted growth of microorganisms. However, the use rate of these chlorine-based antimicrobials is very high because they tend to be rapidly consumed by the high organic load included in both the fruits or vegetables and soil. Further, upon consumption, compounds such as
20 chlorine dioxide decompose producing byproducts such as chlorites and chlorates, while hypochlorite produces trichloromethanes which may be toxic in very low concentrations. Lastly, chlorine dioxide is a toxic gas with an acceptable air concentration limit of 0.1 ppm.
25 Exposure to ClO_2 often leads to headaches, nausea, and respiratory problems, requiring expensive and intricate safety devices and equipment when it is used.

Iodophor antimicrobial agents have also been used for various antimicrobial applications. However,
30 iodophor compounds tend to decompose or may be lost by evaporation when used in an aqueous medium. Thus, long term activity requires a high iodophor concentration.

As a result, a need exists in the food processing industry to provide a means of food transport which also
35 controls soil and microbial load without the use of high concentrations of antimicrobials such as chlorinated compounds and other halogenated constituents.

Summary of the Invention

The invention is a process for preventing microbial growth in aqueous streams comprising the step of
5 applying a percarboxylic acid or a mixture of percarboxylic acids to the aqueous stream.

The process of the invention is unexpectedly effective in preventing the growth of unwanted microorganisms in food transport apparatus. The
10 consumption of peracetic acid is unexpectedly low in view of the organic loading of both fruits or vegetables and microbial soils within the flume water.

The process of the invention provides an antimicrobial agent useful in process water for
15 transporting food products which has a high degree of antimicrobial efficacy and which is safely ingestible by humans while imposing no environmental incompatibility.

Differentiation of antimicrobial "-cidal" or "-static" activity, the definitions which describe the
20 degree of efficacy, and the official laboratory protocols for measuring this efficacy are important considerations for understanding the relevance of antimicrobial agents and compositions. Antimicrobial compositions may effect two kinds of microbial cell
25 damage. The first is a truly lethal, irreversible action resulting in complete microbial cell destruction or incapacitation. The second type of cell damage is reversible, such that if the organism is rendered free or the agent, it can again multiply. The former is
30 termed bacteriocidal and the later, bacteriostatic. A sanitizer and a disinfectant are, by definition, agents which provide antibacterial or bacteriocidal activity. In contrast, a preservative is generally described as an inhibitor or bacteriostatic composition.

35

Brief Description of the Figures

Figure 1 is a graphical depiction of the results of Working Example 4.

Figures 2 and 3 are graphical depictions of the results of Working Example 5.

Figure 4 is a schematic depiction of a flume system used in conjunction with Working Example 7.

Figure 5 is a graphical depiction of certain results obtained from Working Example 7.

Detailed Description of the Invention

A process for preventing microbial growth in aqueous streams, said process comprising the step of applying an effective antimicrobial concentration of a percarboxylic acid composition, to the aqueous stream.

Carboxylic Acid

Among other constituents, the invention comprises a carboxylic acid. Generally, carboxylic acids have the formula $R-COOH$ wherein the R may represent any number of different groups including aliphatic groups, alicyclic groups, aromatic groups, heterocyclic groups, all of which may be saturated or unsaturated as well as substituted or unsubstituted. Carboxylic acids also occur having one, two, three, or more carboxyl groups.

Carboxylic acids have a tendency to acidify aqueous compositions in which they are present as the hydrogen atom of the carboxyl group is active and may appear as an anion. The carboxylic acid constituent within the present composition when combined with aqueous hydrogen peroxide generally functions as an antimicrobial agent as a result of the presence of the active hydrogen atom. Moreover, the carboxylic acid constituent within the invention maintains the composition at an acidic pH.

Carboxylic acids which are generally useful in the process of the invention are those which comprise percarboxylic acids. Percarboxylic acids generally have the formula $R(CO_2H)_N$, where R is an alkyl, arylalkyl,

cycloalkyl, aromatic or heterocyclic group, and N is one or two, or named by prefixing the parent acid with peroxy.

While peroxy carboxylic acids are not very stable, their stability generally increases with increasing molecular weight. Thermal decomposition of these acids may generally proceed by free radical and nonradical paths, by photodecomposition or radical-induced decomposition, or by the action of metal ions or complexes. Percarboxylic acids may be made by the direct, acid catalyzed equilibrium action of 30-98 wt.% hydrogen peroxide with the carboxylic acid, by autoxidation of aldehydes, or from acid chlorides, and hydrides, or carboxylic anhydrides with hydrogen or sodium peroxide.

Percarboxylic acids useful in this invention include peracetic acid, peroctanoic acid, perglutaric acid, persuccinic acid, perdecanoic acid or mixtures thereof. These percarboxylic acids have been found to provide good antimicrobial action with good stability in aqueous streams.

Preferably, the process of the invention uses a combination of peracetic acid and peroctanoic acid. This combination of percarboxylic acids has been found to provide preferred antimicrobial efficacy and stability in the presence of high organic loads. Generally, within the sanitizer, the concentration of peroctanoic acid may range from about 10 wt-% to 90 wt-% and preferably from about 10 wt-% to 20 wt-%. The concentration of peracetic acid may range from about 10 wt-% to 90 wt-% and preferably from about 80 wt-% to 90 wt-%.

In its most preferred mode, the process of the invention uses peracetic acid. Peracetic acid is a peroxy carboxylic acid having the formula:



Generally, peracetic acid is a liquid having an acrid odor and is freely soluble in water, alcohol, ether, and sulfuric acid. Peracetic acid may be prepared through any number of means known to those of skill in the art including preparation from acetaldehyde and oxygen in the presence of cobalt acetate. A 50% solution of peracetic acid may be obtained by combining acetic anhydride, hydrogen peroxide and sulfuric acid. Other methods of formulation of peracetic acid include those disclosed in U.S. Patent No. 2,833,813, which is incorporated herein by reference.

Hydrogen Peroxide

The antimicrobial composition of the invention may also comprise a hydrogen peroxide constituent. Hydrogen peroxide in combination with the percarboxylic acid provides a surprising level of antimicrobial action against microorganisms despite the presence of high loadings of organic sediment. Additionally, hydrogen peroxide may provide an effervescent action which may irrigate any surface to which it is applied. Hydrogen peroxide works with a mechanical flushing action once applied which further cleans the surface of application. An additional advantage of hydrogen peroxide is the food compatibility of this composition upon use and decomposition. For example, combinations of peracetic acid and hydrogen peroxide result in acetic acid, water, and oxygen upon decomposition all of which are food product compatible.

While many oxidizing agents may be used, hydrogen peroxide is generally preferred for a number of reasons. After application of the H_2O_2 /peracetic acid germicidal agent, the residue left merely comprises water and an acidic constituent. Deposition of these products on the surface of application such as a flume, will not adversely effect the process or the food products transported therein.

Hydrogen peroxide (H_2O_2), has a molecular weight of

34.014 and it is a weakly acidic, clear, colorless liquid. The four atoms are covalently bonded in a non-polar H-O-O-H structure. Generally, hydrogen peroxide has a melting point of -0.41°C , a boiling point of 150.2°C , a density at 25°C of $1.4425\text{ grams per cm}^3$, and a viscosity of $1.245\text{ centipoise at }20^{\circ}\text{C}$.

Generally, the concentration of hydrogen peroxide within the composition used in the process of the invention ranges from about 1 weight percent to about 50 weight percent, preferably from about 3 weight percent to about 40 weight percent, and most preferably from about 5 weight percent to about 30 weight percent. This concentration of hydrogen peroxide is most preferred as providing an optimal antimicrobial effect.

These concentrations of hydrogen peroxide may be increased or decreased while still remaining within the scope of the invention.

Adjuvants

The antimicrobial composition of the invention may also comprise any number of adjuvants. Specifically, the composition of the invention may comprise stabilizing agents, wetting agents, as well as pigments or dyes among any number of constituents which may be added to the composition.

Stabilizing agents may be added to the composition of the invention to stabilize the peracid and hydrogen peroxide and prevent the premature oxidation of this constituent within the composition of the invention. Chelating agents or sequestrants generally useful if stabilizing agents in the invention include alkyl diamine polyacetic acid-type chelating agents such as EDTA (ethylene diamine tetraacetate tetrasodium salt), acrylic and polyacrylic acid-type stabilizing agents, phosphonic acid, and phosphonate-type chelating agents among others. Preferable sequestrants include phosphonic acids and phosphonate salts including 1-hydroxy ethyldene-1, 1-diphosphonic acid

($\text{CH}_3\text{C}(\text{PO}_3\text{H}_2)_2\text{OH}$), amino[tri(methylene phosphonic acid)]
([$\text{CH}_2\text{PO}_3\text{H}_2$]₂(ethylene diamine[tetra methylene-phosphonic
acid])), 2-phosphene butane-1, 2, 4-tricarboxylic acid,
as well as the alkyl metal salts, ammonium salts, or
5 alkyloyl amine salts, such as mono, di, or tetra-
ethanolamine salts. The stabilizing agent is used in a
concentration ranging from about 0 weight percent to
about 20 weight percent of the composition, preferably
from about 0.1 weight percent to about 10 weight percent
10 of the composition, and most preferably from about 0.2
weight percent to 5 weight percent of the composition.

Also useful in the composition of the invention are
wetting and defoaming agents. Wetting agents function
to increase the penetration activity of the
15 antimicrobial composition of the invention. Wetting
agents which may be used in the composition of the
invention include any of those constituents known within
the art to raise the surface activity of the composition
of the invention.

20 Along these lines surfactants, and especially
nonionic surfactants, may also be useful in the present
invention. Nonionic surfactants which may be useful in
the present invention are those which comprise ethylene
oxide moieties, propylene oxide moieties, as well a
25 mixtures thereof, and ethylene oxide-propylene oxide
moieties in either heteric or block formation.
Additionally useful in the present invention are
nonionic surfactants which comprise an alkyl ethylene
oxide compounds, alkyl propylene oxide compounds, as
30 well as mixtures thereof, and alkyl ethylene oxide-
propylene oxide compounds where the ethylene oxide
propylene oxide moiety is either in heteric or block
formation. Further useful in the present invention are
nonionic surfactants having any mixture or combination
35 of ethylene oxide-propylene oxide moieties linked to a
alkyl chain where the ethylene oxide and propylene oxide
moieties may be in any randomized or ordered pattern and

of any specific length. Nonionic surfactants useful in the present invention may also comprise randomized sections of block and heteric ethylene oxide propylene oxide, or ethylene oxide-propylene oxide.

5 Generally, the concentration of nonionic surfactant used in the invention may range from about 0 wt-% to about 5 wt-% of the composition, preferably from about 0 wt-% to about 2 wt-% of the concentrate composition, and most preferably from about 0 wt-% to about 1 wt-% of the
10 composition.

 The composition used in the process of the invention may also contain additional ingredients as necessary to assist in defoaming.

 Generally, defoamers which may be used in accordance
15 with the invention include silica and silicones; aliphatic acids or esters; alcohols; sulfates or sulfonates; amines or amides; halogenated compounds such as fluorochlorohydrocarbons; vegetable oils, waxes, mineral oils as well as their sulfated derivatives;
20 fatty acid soaps such as alkali, alkaline earth metal soaps; and phosphates and phosphate esters such as alkyl and alkaline diphosphates, and tributyl phosphates among others; and mixtures thereof.

 Especially preferable, are those antifoaming agents
25 or defoamers which are of food grade quality given the application of the process of the invention. To this end, one of the more effective antifoaming agents comprises silicones. Silicones such as dimethyl silicone, glycol polysiloxane, methylphenol
30 polysiloxane, trialkyl or tetraalkyl silanes, hydrophobic silica defoamers and mixtures thereof may all be used in defoaming applications. Commercial defoamers commonly available include silicones such as Ardefoam® from Armour Industrial Chemical Company which is a silicone
35 bound in an organic emulsion; Foam Kill® or Kresseo® available from Krusable Chemical Company which are silicone and non-silicone type defoamers as well as

silicone esters; and Anti-Foam A® and DC-200™ from Dow Corning Corporation which are both food grade type silicones among others. These defoamers are generally present at a concentration range from about 0 wt-% to 5 wt-%, preferably from about 0 wt-% to 2 wt-%, and most preferably from about 0 wt-% to about 1 wt-%.

The invention may also contain any number of other constituents as necessitated by the application, which are known to those of skill in the art and which may facilitate the activity of the present invention.

The composition used in the invention may comprise:

<u>Composition (Wt-%)</u>				
	<u>Useful</u>	<u>Working</u>	<u>Preferred</u>	
15	Percarboxylic Acid	2-25	2-20	4-20
	H ₂ O ₂	1-45	5-35	7-30
	Acetic Acid	1-70	3-55	5-45
	Water	Balance	Balance	Balance

<u>Initial Concentration in Transport Water</u>				
	<u>Useful</u>	<u>Working</u>	<u>Preferred</u>	
20				
	<u>Constituent</u>			
25	Percarboxylic Acid	5-100 ppm	5-60 ppm	10-50 ppm
	H ₂ O ₂	5-500 ppm	5-300 ppm	5-250 ppm

Once the antimicrobial of the invention is applied to any given transport stream, the antimicrobial will be subjected to a demand resulting from microbes present in the stream as well as other organic or inorganic material present in the stream. As a general guideline, not limiting of the invention, the following concentrations of antimicrobial may be found after demand.

35

		<u>Residual Concentration (ppm)</u> <u>After Demand</u>		
	<u>Constituent</u>	<u>Useful</u>	<u>Working</u>	<u>Preferred</u>
5	Percarboxylic Acid	1-85	1-45	1-35
	H ₂ O ₂	1-490	1-290	1-240

While the demand may reduce the antimicrobial concentration to zero, at least about 1 ppm of POAA is generally preferred to provide the intended efficacy.

10

Generation of Peroxy Acids

The process of the invention may also be initiated through the use of peroxy acid concentrate compositions. In such a case, the percarboxylic acid may either be generated naturally or through the combination of a hydrogen peroxide concentrate together with a carboxylic acid concentrate at the sight of use such as that process which is disclosed in Lokkesmoe et al, U.S. Patent No. 5,122,538, issued June 16, 1992, which is incorporated herein by reference. In such a case, the composition may be formed from a hydrogen peroxide concentrate comprising varying levels of hydrogen peroxide and stabilizer as shown in the table below.

		<u>Concentration (Wt-%)</u>		
25	<u>Constituent</u>	<u>Useful</u>	<u>Working</u>	<u>Preferred</u>
	Hydrogen Peroxide	5-70	15-70	25-60
	Stabilizer	0-10	0-5	0.1-3
	H ₂ O	20-95	25-85	37-75

When combined with a carboxylic acid, the two concentrates result in a peroxy carboxylic acid. Generally, the carboxylic acid concentrate comprises a carboxylic acid in water as shown in the table found below.

		<u>Concentration (Wt-%)</u>		
35	<u>Constituent</u>	<u>Useful</u>	<u>Working</u>	<u>Preferred</u>
	Carboxylic Acid	50-100	65-100	80-100
	Water	0-50	0-35	0-20

Working Examples

The invention will now be described in more detail by reference to the following examples. The only proper construction of these examples is as non-limiting illustrative examples showing various formulations, stabilities, and applications of the invention.

Working Example 1

To prepare stock solution of concentrate peracetic acid (or "POAA") formula for use in the flume experiments, the following components were combined.

	<u>Component</u>	<u>Wt-%</u>
	Acetic Acid	43.85
	Hydrogen Peroxide 35%	50.85
15	Dequest 2010 (60% active)	1.5
	1-Hydroxyethylidene-1,1-Diphosphonic Acid	
	H ₂ O	3.8

The result of this combination was a composition having the following constituency.

		<u>Wt-%</u>
	Acetic Acid	32.0
25	H ₂ O ₂	11.1
	Request 2010	0.90
	H ₂ O	41.0
	Peracetic Acid	15.0

30

Working Example 2

In the second working example, the immediate demand of 1% and 3% tomato solutions for POAA was determined. POAA in pure water (control) was compared with similar dilutions in 1% and 3% tomato solutions.

Table 1

	<u>Control</u> <u>(no tomatoes)</u>	<u>1% Tomato</u>	<u>3% Tomato</u>
40	Mean POAA conc. (ppm)	111.8	112.25
	Mean H ₂ O ₂ conc. (ppm)	65.2	65.3
	Number of Trials	3	2
	Std Dev. (POAA)	0.61	0.21
45	Std Dev. (H ₂ O ₂)	0.10	0.14

15

No initial decrease in the POAA concentration was seen.

Working Example 3

The third working example expanded the time frame of testing to 8 days for 20 ppm POAA solutions made from both 5% and 15% POAA formulations. Again, 1% tomato solutions were used. Tables 2-5 show that 93% to 100% of the POAA remains initially. After four days, 32% to 61% POAA was left after contacting a 1% tomato solution for the 5% and 15% POAA formulas, respectively. Tables 4 and 5 represent hydrogen peroxide control solutions compared under the same analytical protocol.

Table 2

(5% POAA)

DAY	<u>CONTROL</u>		<u>1% TOMATO</u>	
	<u>POAA PPM</u>	<u>%REMAIN</u>	<u>POAA PPM</u>	<u>%REMAIN</u>
0	17.66	100%	16.47	100%
1	19.66	111%	13.23	80%
20 2	16.04	91%	10.23	62%
3	16.04	91%	7.22	44%
4	--	--	5.24	32%
5	--	--	--	--
6	--	--	--	--
25 7	13.38	76%	--	--
8	12.77	72%	--	--

pH=4.01

pH=4.60

30 INITIAL PERCENTAGE OF CONTROL = 93%

Table 3

(15% POAA)

DAY	<u>CONTROL</u>		<u>1% TOMATO</u>	
	<u>POAA PPM</u>	<u>%REMAIN</u>	<u>POAA PPM</u>	<u>%REMAIN</u>
0	17.19	100%	18.87	100%
1	17.04	99%	15.86	84%
40 2	15.20	88%	14.71	78%
3	13.76	80%	13.00	69%
4	--	--	11.55	61%
5	--	--	--	--
6	--	--	--	--
45 7	7.90	46%	--	--
8	6.99	41%	7.30	39%

pH=3.92

pH=4.13

INITIAL PERCENTAGE OF CONTROL = 110%

16

Table 4

(5% POAA)

		<u>CONTROL</u>		<u>1% TOMATO</u>	
	<u>DAY</u>	<u>H2O2 PPM</u>	<u>%REMAIN</u>	<u>H2O2 PPM</u>	<u>%REMAIN</u>
5	0	83.45	100%	82.27	100%
	1	84.69	101%	81.54	99%
	2	83.77	100%	80.89	98%
	3	84.32	101%	79.53	97%
	4	--	--	75.85	92%
10	5	--	--	--	--
	6	--	--	--	--
	7	84.35	101%	--	--
	8	84.66	101%	--	--
pH=4.01			pH=4.60		

15 INITIAL PERCENTAGE OF CONTROL = 99%

Table 5

(15% POAA)

	<u>CONTROL</u>		<u>1% TOMATO</u>		
<u>DAY</u>	<u>H2O2 PPM</u>	<u>%REMAIN</u>	<u>H2O2 PPM</u>	<u>%REMAIN</u>	
20	0	10.27	100%	10.89	100%
	1	10.99	107%	10.40	96%
	2	11.15	109%	10.64	98%
	3	11.15	109%	10.23	94%
25	4	--	--	10.00	92%
	5	--	--	--	--
	6	--	--	--	--
	7	10.18	99%	--	--
	8	10.57	103%	8.06	74%
30	pH=3.92		pH=4.13		

INITIAL PERCENTAGE OF CONTROL = 106%

35 Working Example 4

The fourth working example provided a set of stability experiments expanded to the use of peas, beans, and corn. Tables 6-13 show the stability of a formula which is mostly peracetic acid (with peroctanoic acid being approximately 10 wt-% of the total peracetic and peroctanoic acid content) in 1% solutions of these vegetables. The initial concentrations were 70%, 100% and 90% of the control solutions (with no vegetable) for corn, beans, and peas, respectively. After 3 days, 31%, 47% and 32%, respectively, of the initial concentration of peracids such as POAA was left for these vegetables.

The peracids showed surprising stability in solutions comprising a high concentration of organic material.

Table 6

(CONTROL)

	TOTAL PERACIDS (AS POAA)		
	<u>DAY</u>	<u>PPM</u>	<u>%REMAIN</u>
	0	18.85	100%
	1	19.76	100%
10	2	18.77	100%
	3	16.80	89%

Table 7

(1% CORN)

	TOTAL PERACIDS (AS POAA)		
	<u>DAY</u>	<u>PPM</u>	<u>%REMAIN</u>
	0	13.15	100%
	1	8.51	65%
25	2	6.16	47%
	3	4.03	31%

INITIAL PERCENTAGE OF CONTROL = 70%

Table 8

(1% BEANS)

	TOTAL PERACIDS (AS POAA)		
	<u>DAY</u>	<u>PPM</u>	<u>%REMAIN</u>
	0	21.36	100%
	1	17.48	82%
40	2	14.36	67%
	3	9.96	47%

50 INITIAL PERCENTAGE OF CONTROL = 113%

18

Table 9

(1% PEAS)

5		TOTAL PERACIDS (AS POAA)	
	<u>DAY</u>	<u>PPM</u>	<u>%REMAIN</u>
10	0	18.09	100%
	1	12.46	69%
	2	10.41	58%
	3	5.70	32%

INITIAL PERCENTAGE OF CONTROL = 96%

Table 10

(CONTROL)

	<u>DAY</u>	<u>H2O2 PPM</u>	<u>%REMAIN</u>
25	0	10.30	100%
	1	10.98	107%
	2	10.91	106%
	3	10.85	105%

Table 11

(1% CORN)

35	<u>DAY</u>	<u>H2O2 PPM</u>	<u>%REMAIN</u>
	0	15.67	100%
	1	7.21	46%
	2	5.71	36%
40	3	1.70	11%

INITIAL PERCENTAGE OF CONTROL = 152%

Table 12

(1% BEANS)

	<u>DAY</u>	<u>H2O2 PPM</u>	<u>%REMAIN</u>
50	0	8.84	100%
	1	3.09	35%
	2	1.63	18%
	3	1.09	12%

INITIAL PERCENTAGE OF CONTROL = 86%

Table 13

(1% PEAS)

	<u>DAY</u>	<u>H2O2 PPM</u>	<u>%REMAIN</u>
	0	8.57	100%
10	1	4.83	56%
	2	3.37	39%
	3	0.78	9%

INITIAL PERCENTAGE OF CONTROL = 83%

Working Example 5

Experiments testing the efficacy of POAA on molds and bacteria showed no microbial growth at concentrations of 5, 10 and 20 ppm POAA in 1% peas solution. As can be seen in Figure 1, later experiments show good control of molds with 10-30 ppm POAA in 1% peas solution.

Working Example 6

An experiment was completed to obtain a better understanding of the effect of catalase on POAA/H₂O₂ solutions. In this experiment, POAA and H₂O₂ were added to solutions containing a variety of concentration of catalase and in one a 1:50 dilution of an overnight culture of S.aureus. POAA and H₂O₂ concentrations were then monitored in the presence of catalase by titration along with the catalase activity in each solution. The results of this experiment are shown in Table 14 and in Figures 2 and 3.

Table 14POAA Stability in the Presence of Catalase (CTS)

	<u>Sample</u>	<u>*Catalase Activity</u>	<u>Initial ppm POAA H₂O₂</u>	
40	Blank (PO4)	0	100	277
	S.aureus	2.8	158	505
	0.5 U/ml	0.34	65	185
45	1.0 U/ml	0.66	95	225
	20 U/ml	20	95	155

Table 14 (Cont'd.)

	<u>Sample</u>	<u>*Catalase Activity</u>	<u>1 Hour ppm</u>	
			<u>POAA</u>	<u>H₂O₂</u>
5	Blank (PO4)	0	82	268
	S.aureus	1.1	158	27
	0.5 U/ml	ND**	68	102
	1.0 U/ml	ND	76	71
10	20 U/ml	10	65	4
	<u>Sample</u>	<u>*Catalase Activity</u>	<u>4 Hours ppm</u>	
			<u>POAA</u>	<u>H₂O₂</u>
15	Blank (PO4)	0	65	267
	S.aureus	0.08	131	8
	0.5 U/ml	ND	65	82
	1.0 U/ml	ND	57	66
20	20 U/ml	.21	52	7

*Activity is expressed as micromoles of H₂O₂ hydrolyzed in 1 minute.

**Not Detected

25 The test was done at room temperature and the concentrations of catalase used were lower than those seen in a culture. The presence of the catalase did accelerate the decomposition of H₂O₂ but did not accelerate the decomposition of the POAA at these

30 concentrations.

Working Example 7

An analysis of the invention was then undertaken in the context of an actual flume delivery system. As can be seen in Figure 4, a flume system comprising a make-up tank 10, a flow line 11, a flume tank 12, an overflow tank 14 with discharge pipe or drain 13, pumpline 15, pump 16, and recycle line 18 were assembled to model the conditions in food transport flumes used in food processing plants. The make-up water comprised 16 grains/gal of CaCO₃ and was introduced into the flume at a rate of 343 ml/min. A pea solution was introduced into the make-up tank 10 comprising 10% ground peas in hard water. The pea solution was diluted to 1% in the flume through a flow rate of 42.5 ml/min. Also added to

45 the make-up water was a dirt solution comprising 3.6%

top soil which was diluted to 0.3% in the flume by a flow rate of 35 ml/min. Lastly, a sanitizer was added to the flume assembly and diluted by a factor of 100 through a flow rate of 42 ml/min. The initial concentration and formulation for the sanitizers analyzed can be seen in Table 15 below.

Table 15

	<u>Working Example</u>	<u>Active/ Concentration</u>	<u>Condition</u>
10	7A	30 ppm POAA	Sterile peas
	Control 1	--	Sterile peas
	7B	NaOCl/110 ppm Cl	--
15	7C	NaOCl/30 ppm Cl	--
	7D	30 ppm POAA	Pulsed feed
	7E	40 ppm POAA	Continuous feed
	7F	3 ppm POOA*/ 27 ppm POAA	--
20	Control 2	--	--
	7G	20 ppm ClO ₂	3.7 ml/min flow rate
	7H	1.5 ppm POOA*/ 13.5 POAA	--

25 *POOA is Peroctanoic Acid

The total flow rate in the flume was 425 ml/min with a recycle flow rate created by pump 16 of 3 gallons/min. The total flume volume was 2.25 gallons with overflow discharged out of overflow tank 14 into discharge reservoir 13. Analysis for metals present in the water exiting the flume from a previous experiment gave an average of 13.4 ppm iron (Fe), 0.28 ppm copper (Cu), and 0.52 ppm manganese (Mn).

35 The results of the analysis can be seen in Table 16 and Figure 5.

Table 16

Working Example	Avg. Residual (ppm)	Avg. Demand (ppm)	Avg. pH	Avg. Flume cts.	Avg. Log Red. vs Control	Avg. Pea cts.
10 7A	14.9	15.1	7.5	1.7E+03	2.8	5.0E+04
Control 1	--	--	NA	1.0E+06		3.9E+04
15 7B	41	69	8.4	2.2E+04	2.0	1.2E+06
7C	19	11	8.2	4.9E+04	1.7	1.2E+06
7D	15.7	14.3	7.53	9.5E+03	2.4	2.0E+06
20 7E	24.1	15.9	7.4	1.5E+04	2.2	7.1E+04
7F	20	10	6.3	9.5E+03	2.4	2.3E+04
Control 2	--	--	NA	2.2E+06	--	1.4E+05
25 7G	0	17.5	NA	1.4E+04	2.2	1.7E+05
7H	5	10	7.2	4.0E+04	1.7	5.0E+05

30

The above discussion, examples, and data illustrate our current understanding of the invention. However, since many variations of the invention can be made without departing from the spirit and scope of the invention, the invention resides wholly in the claims hereinafter appended.

WE CLAIM:

1. A method of controlling microbial growth in aqueous streams comprising the step of treating the aqueous stream used for delivery of food products with an effective antimicrobial amount of percarboxylic acid.
2. The method of claim 1 wherein said percarboxylic acid comprises peracetic acid.
3. The method of claim 1 wherein said percarboxylic acid comprises a C₂-C₁₂ percarboxylic acid.
4. The method of claim 3 wherein said percarboxylic acid is selected from the group consisting of peracetic, peroctanoic, perdecanoic, persuccinic, perglutaric, or mixtures thereof.
5. The method of claim 1 additionally comprising hydrogen peroxide.
6. The method of claim 5 wherein said hydrogen peroxide is present in an initial concentration ranging from about 5 ppm to 500 ppm in the aqueous stream.
7. The method of claim 6 wherein said percarboxylic acid comprises peracetic acid present in an initial concentration ranging from about 5 ppm to 100 ppm in the aqueous stream.
8. The method of claim 6 wherein said percarboxylic acid is selected from the group consisting of peracetic acid, peroctanoic acid, or mixtures thereof present in a total concentration ranging from about 5 ppm to 100 ppm.
9. The treated aqueous food delivery stream resulting from the method of claim 1.
10. A method of controlling microbial growth in aqueous streams by administering an antimicrobial composition to the stream, said antimicrobial composition comprising:
 - (i) from about 5 to 100 ppm percarboxylic acid initially in the transport stream; and
 - (ii) from about 5 to 500 ppm hydrogen peroxide initially in the transport stream,

said method comprising the steps of

- (a) forming said antimicrobial composition by combining a first concentrate comprising hydrogen peroxide and a second concentrate comprising a carboxylic acid; and
- (b) administering said antimicrobial composition to said aqueous stream used for the delivery of food products.

11. The method of claim 10 wherein said percarboxylic acid comprises peracetic acid.

12. The method of claim 10 wherein said percarboxylic acid comprises a C₂-C₁₂ percarboxylic acid.

13. The method of claim 10 wherein said percarboxylic acid is selected from the group consisting of peracetic, peroctanoic, perdecanoic, persuccinic, perglutaric, or mixtures thereof.

14. The method of claim 11 wherein said peracetic acid is present in a concentration ranging from about 5 to 100 ppm in the food delivery stream.

15. The method of claim 10 wherein said percarboxylic acid is selected from the group consisting of peracetic acid, peroctanoic acid, or mixtures thereof present in an initial concentration ranging from about 5 ppm to 100 ppm.

16. The method of claim 10 wherein said antimicrobial composition comprises a wetting agent present in a concentration ranging from about 0 to 1 wt-%.

17. The treated aqueous food delivery stream resulting from the method of claim 10.

18. A method of controlling microbial growth in aqueous streams, said method comprising the step of treating said aqueous stream used for the transport of food products with an antimicrobial agent, said antimicrobial composition comprising:

- (a) from about 5 to 100 ppm peracetic acid initially in the transport stream; and

(b) from about 5 to 500 ppm hydrogen peroxide initially in the transport stream.

19. The method of claim 18 wherein said antimicrobial composition comprises a nonionic
5 surfactant.

20. The method of claim 18 wherein said peracetic acid is present in a concentration ranging from about 5 to 60 ppm in the food delivery stream.

21. The method of claim 18 wherein said hydrogen
10 peroxide is present in a concentration ranging from about 5 to 300 ppm.

22. The treated aqueous food delivery stream resulting from the method of claim 18.

23. A method of controlling microbial growth in
15 aqueous streams, said method comprising the steps of:

(a) forming an antimicrobial composition by combining a first concentrate comprising from about 5 wt-% to 70 wt-% H_2O_2 with a second concentrate comprising from about 50 wt-% to 100 wt-% carboxylic
20 acid;

(b) forming a percarboxylic acid within said antimicrobial composition; and

(c) treating the aqueous stream used for delivery of food with said antimicrobial
25 composition.

24. The method of claim 23 wherein said percarboxylic acid comprises peracetic acid.

25. The method of claim 23 wherein said percarboxylic acid comprises a C_2 - C_{12} percarboxylic acid.

30 26. The method of claim 25 wherein said percarboxylic acid is selected from the group consisting of peracetic, peroctanoic, perdecanoic, persuccinic, perglutaric, or mixtures thereof.

27. The method of claim 23 wherein said hydrogen
35 peroxide is present in an initial concentration ranging from about 5 ppm to 500 ppm in the treated aqueous food delivery stream.

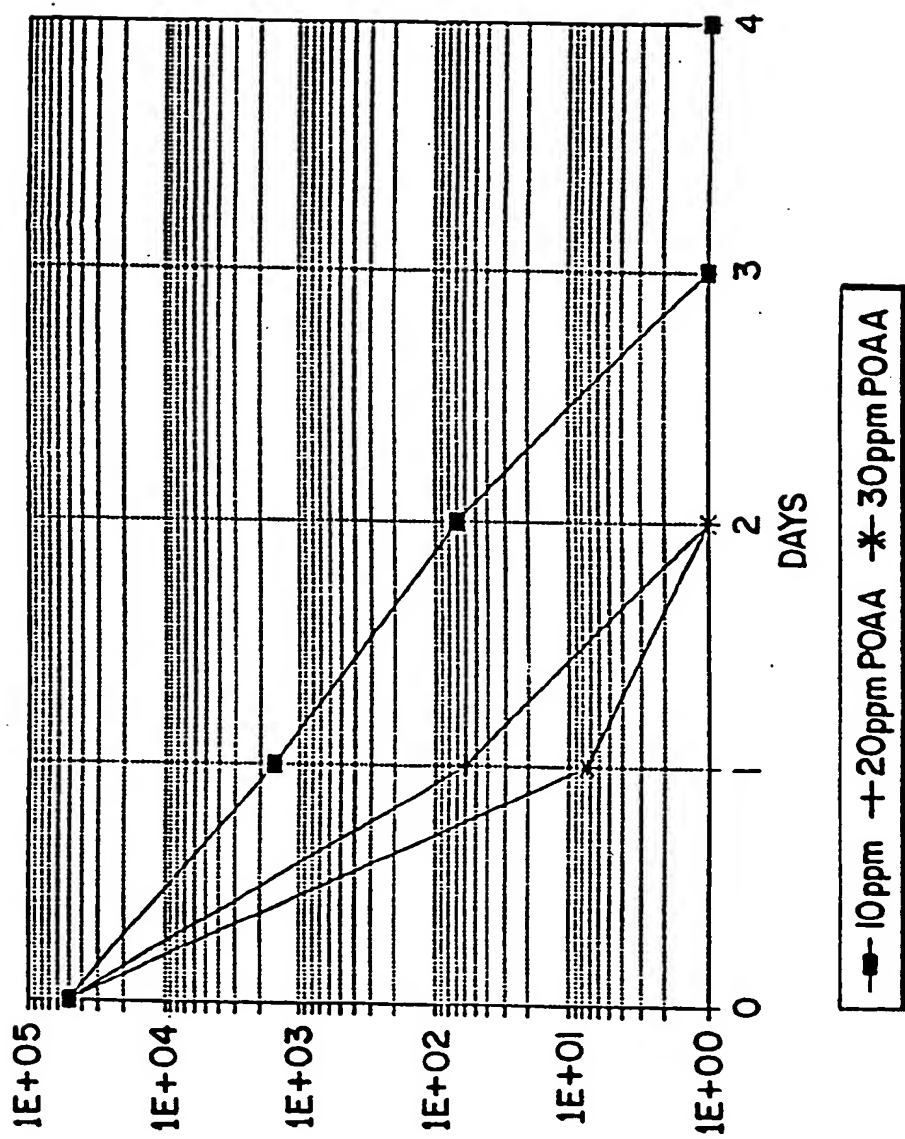
28. The method of claim 23 wherein said percarboxylic acid comprises peracetic acid present in a concentration ranging from about 5 ppm to 100 ppm in the aqueous food delivery stream.

5 29. The method of claim 23 wherein said percarboxylic acid is selected from the group consisting of peracetic acid, peroctanoic acid, or mixtures thereof present in a total concentration ranging from about 5 ppm to 100 ppm.

10 30. The treated aqueous food delivery stream resulting from the method of claim 23.

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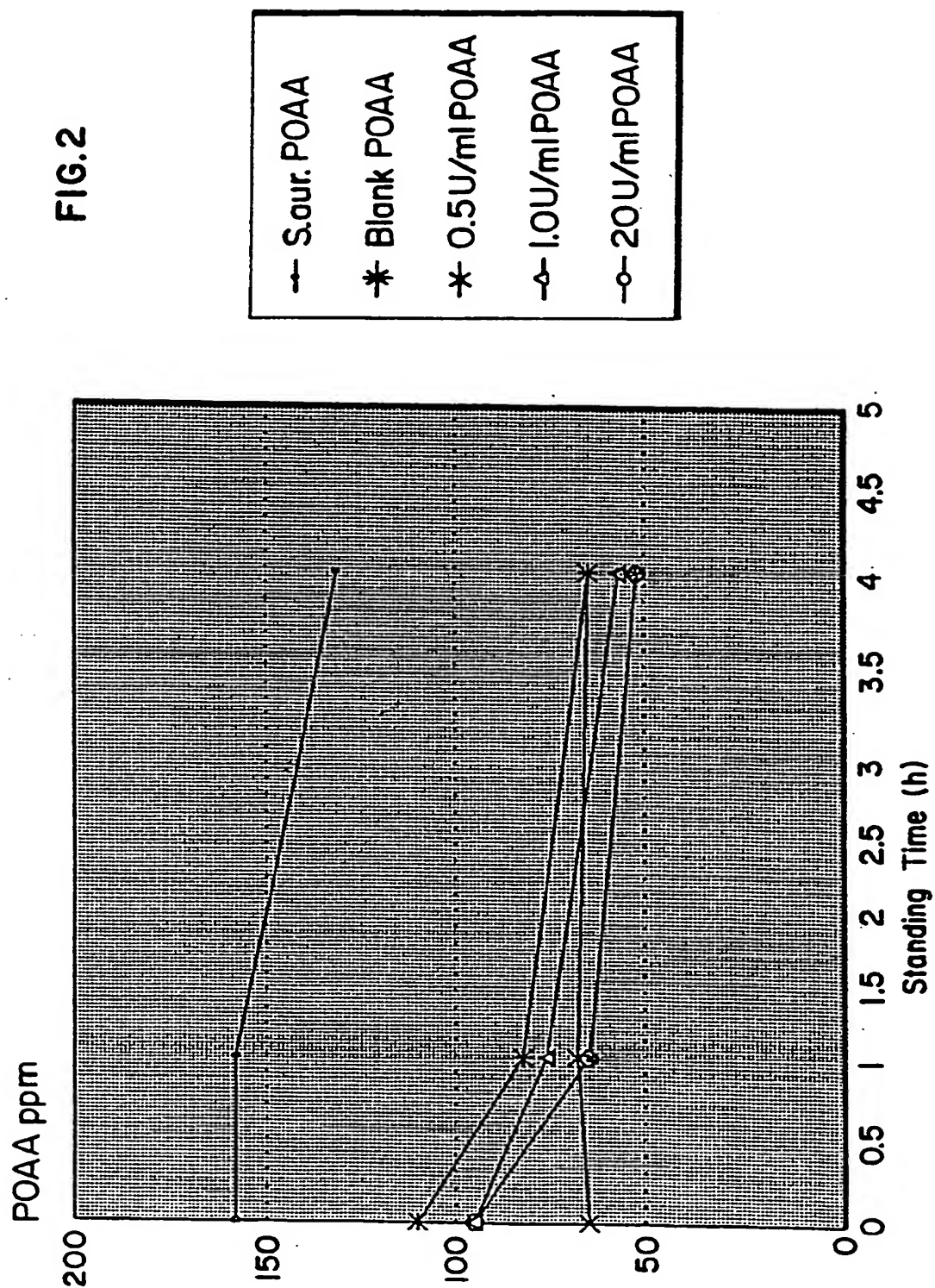
FIG. 1



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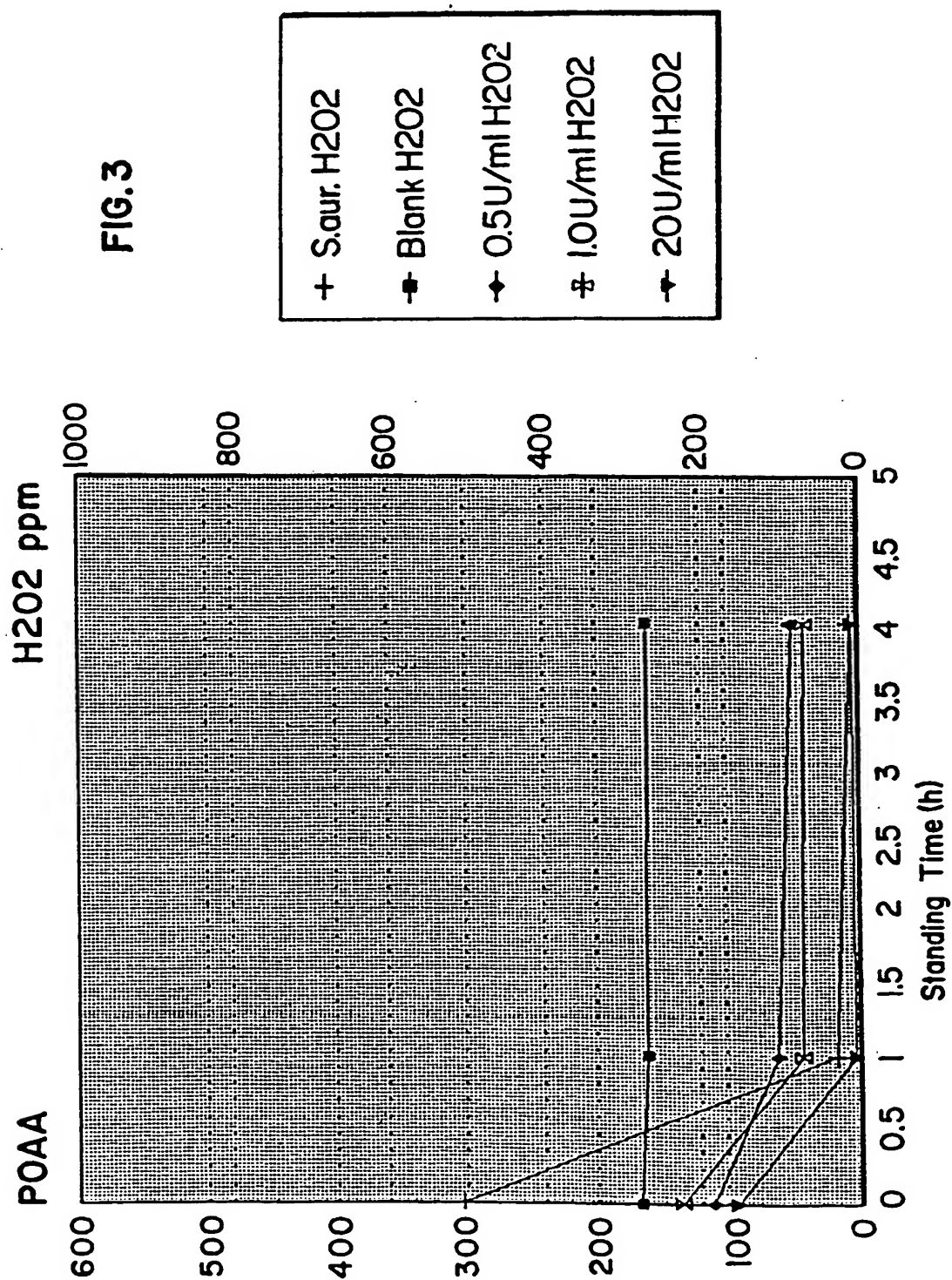
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FIG. 2



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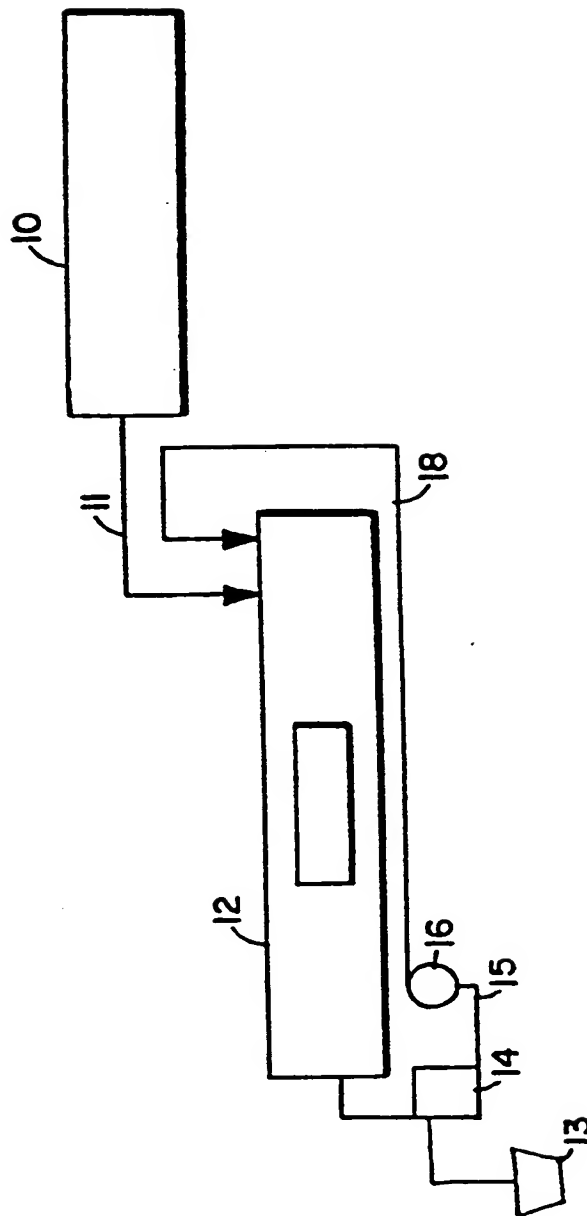
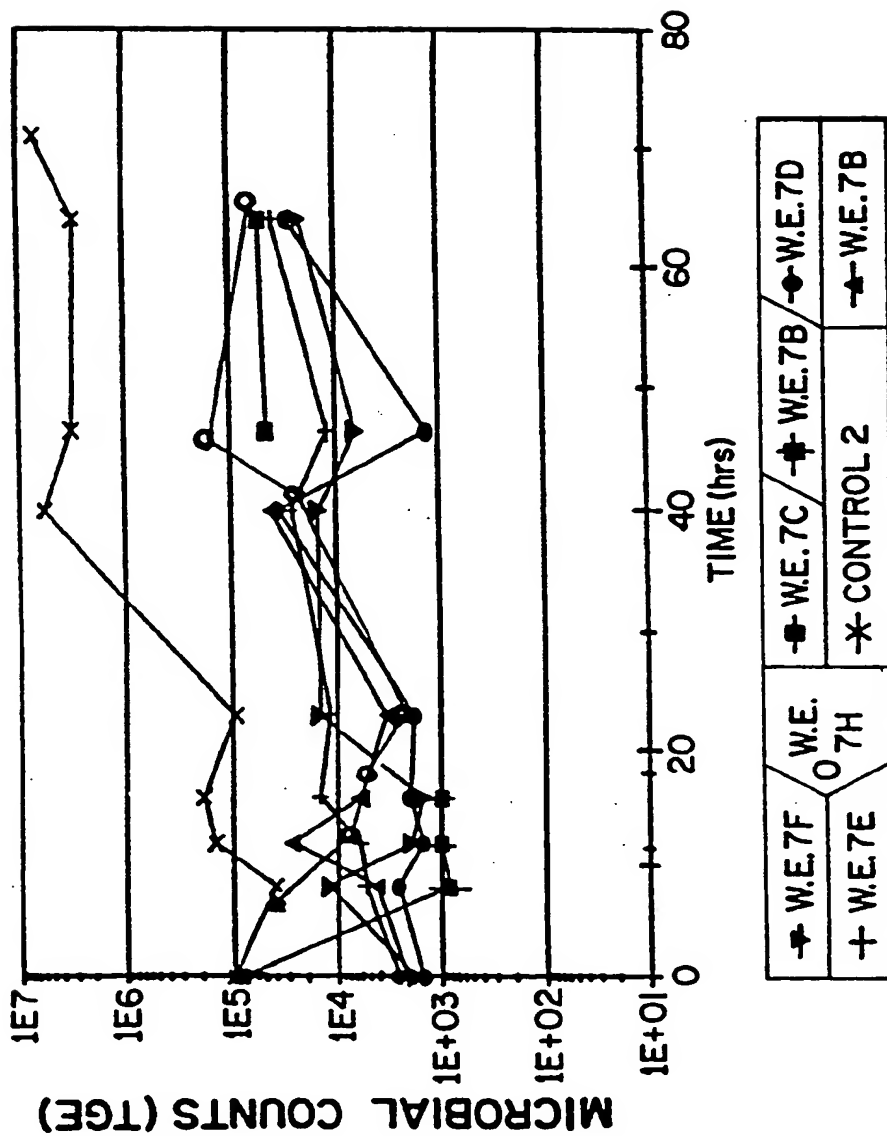


FIG. 4

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FIG. 5



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INTERNATIONAL SEARCH REPORT

Intern. Appl. No.
PCT/US 93/07952

A. CLASSIFICATION OF SUBJECT MATTER
IPC 5 A01N37/16 C02F1/72

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 5 A01N C02F

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	WO,A,93 02973 (W. TRAWOGER ET AL.) 18 February 1993 see claims ---	1-30
Y	EP,A,0 361 955 (ALBRIGHT & WILSON) 4 April 1990 see claims see page 2, line 24 - line 32 see page 3, line 43 - line 45 ---	1-30
A	S.S. BLOCK 'disinfection, sterilisation and preservation' 1991, LEA & FEBIGER, PHILADELPHIA cited in the application chapter 9 see page 179, column 1, line 13 - line 14 --- -/--	1-30

☒ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

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Date of the actual completion of the international search

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INTERNATIONAL SEARCH REPORT

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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	<p>GB,A,2 257 630 (INTEROX CHEMICALS) 20 January 1993 see page 11, line 17 - page 12, line 3 see page 12, line 26 - line 28 -----</p>	1-30

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INTERNATIONAL SEARCH REPORT

Information on patent family members

Intern. Appl. Application No

PCT/US 93/07952

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		AU-A- 4179689	05-04-90
		GB-A, B 2224441	09-05-90
		US-A- 5168655	08-12-92
GB-A-2257630	20-01-93	NONE	

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